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CARDIORESPIRATORY EFFECTS OF VATINOXAN IN BLESBOK (*DAMALISCUS PYGARGUS PHILLIPSI*) IMMOBILIZED WITH THIAFENTANIL—MEDETOMIDINE

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Abstract: Combinations of a low dose of opioid, such as thiafentanil, and a high dose of medetomidine, are increasingly being used for immobilization of African ungulates. Both drugs can have undesirable cardiorespiratory effects. In this study we assessed whether vatinoxan, a peripherally acting alpha$_2$–adrenergic receptor antagonist, can be used to alleviate some of these effects without affecting the immobilization quality. Eight healthy, female, boma–confined blesbok (*Damaliscus pygargus phillipsi*), weighing a mean (SDtion) of 56.8 (4.4) kg, were immobilized twice in a randomized cross–over study with a 2–wk washout period using (1) 0.5 mg thiafentanil + 1.5 mg medetomidine (TM), (2) TM + vatinoxan: 0.5 mg thiafentanil + 1.5 mg medetomidine + 15 mg vatinoxan per milligram medetomidine (total of 22.5 mg, administered intramuscularly at 10 min post recumbency). Heart rate, respiratory rate, rectal temperature, oxygen saturation (SpO_2), arterial blood pressure, and sedation scores from 1 to 5 (1 = limited effect; 5 = excessively deep) were measured every 5 min. Arterial blood gases (PaO_2 and $PaCO_2$) were measured at 10, 15, 25, and 35 min postrecumbency and the alveolar—arterial oxygen gradient ($P[A–a]O_2$) was calculated. Induction times and immobilization quality did not differ between groups. The heart rate was significantly higher and the mean arterial pressure significantly lower in blesbok after receiving vatinoxan. All animals were hypoxemic and there were no significant differences in the respiratory rates, PaO_2, $PaCO_2$, SpO_2, or $P(A–a)O_2$ gradients at any time point. Although vatinoxan did not improve respiratory variables and blood oxygenation in these animals, the change in cardiovascular variables may suggest that it improves tissue perfusion, a positive outcome that requires further investigation.

INTRODUCTION

Alpha$_2$–adrenergic receptor agonists (alpha$_2$–agonists) such as medetomidine are widely used in domestic and nondomestic animals, either for sedation when used as a sole agent, or for the potentiation of immobilization when used in combination with cyclohexanes or opioids.[17] Alpha$_2$–agonists produce dose–dependent sedation, muscle relaxation, and analgesic effects by stimulating alpha$_2$–adrenergic receptors that exist pre– and post–synaptically in tissues throughout the body.[27] Cardiorespiratory side effects of alpha$_2$–agonists are highly dose and species dependent. Cardiovascular effects include an initial increase in blood pressure and reflex bradycardia because of peripheral vasoconstriction followed by centrally mediated reduction in sympathetic tone, heart rate, blood pressure and cardia output.[27] Respiratory effects may include respiratory depression, reduced response to carbon dioxide tension, and hypoxemia and hypercapnia, especially when administered with opioids or benzodiazepines.[21,34]

Dexmedetomidine and medetomidine are considered the most selective of all the alpha$_2$–agonists[27] and are available in highly concentrated formulations that make them suitable for inclusion in projectile darting systems used in zoo and wild animals. In recent years, immobilization protocols containing a low dose of a potent opioid, such as thiafentanil, and a high dose of medetomidine has gained popularity for use in African wildlife.[20] Compared to the traditionally used higher–dose potent opioid protocols[20] there is a little published information regarding the physiological effects of these combinations nor how such effects potentially could be mitigated.

One alpha$_2$–adrenergic receptor antagonist (alpha$_2$ antagonists), vatinoxan, has gained interest as a potential tool for mitigating adverse effects of medetomidine and dexmedetomidine. Vatinoxan acts preferentially on the peripherally located alpha$_2$–adrenergic receptors[14] and, as a result of its poor lipid solubility, only minimally crosses

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the blood—brain barrier. Its direct effect is therefore limited to peripheral tissues.[4] When co–administered with alpha$_2$–agonists, it abates the initial vasoconstriction and the consequent hemodynamic disturbances induced by an alpha$_2$–agonist without affecting the centrally mediated desirable sedative effects.[14] Its effects appear to be dose–dependent, and it has been shown to be effective in a number of domestic species.[1,2,13,16,33] More recently, it has been investigated for its potential benefits in wildlife species, including Patagonian maras (*Dolichotis patagonum*),[9] wild boars (*Sus scrofa*),[6] red deer (*Cervus elaphus*),[7] and markhors (*Capra falconeri*).[32]

The aim of this study was to investigate the effect of vatinoxan on the cardiorespiratory response of blesbok (*Damaliscus pygargus phillipsi*) immobilized with a low dose of thiafentanil combined with medetomidine. We hypothesized that the administration of vatinoxan would lead to higher heart rate, partial pressure of oxygen (PaO_2), and alveolar—arterial oxygen gradient ($P[A–a]O_2$), as well as lower mean arterial blood pressure (MAP) and partial pressure of carbon dioxide ($PaCO_2$) compared to when animals did not receive vatinoxan.

MATERIALS AND METHODS

The study received ethical approval from both Wildlife Pharmaceutical's Animals Ethics Committee (approval WPAEC–2022–VATINOXAN–51–B) and the University of Pretoria's Animal Ethics Committee (approval REC083–21). Eight adult, female blesbok (mean = 56.8 kg; SD = 4.4 kg) originating from local game farms were translocated to the Wildlife Pharmaceutical's Wildlife Research Facility (25°31′25.2″S, 31°06′50.8″E) and acclimatized in enclosures (6 × 8 m) for 4 wk. Selection criteria for the animals included being female, non–pregnant, and in apparent good health. Exact ages were not known, but estimated to range from approximately 3 to 8 yr. On arrival, all the animals were immobilized and weighed, the horns were piped and marked with white tape for identification, and each animal underwent a general health check. They were provided feed and water ad libitum and were allowed to move between enclosures in between trials.

The study was conducted in two– phases. First, a pilot study was conducted in which the appropriate dose of vatinoxan (Vetcare LtdP, Mäntsälä, 04600, Finland) in blesbok was established. Three blesbok were immobilized with thiafentanil (10 mg/ml, Wildlife Pharmaceuticals (Pty) Ltd, White River 1240, South Africa; 0.01mg/kg) + medetomidine (10 mg/ml, Kyron Laboratories (Pty) Ltd, Johannesburg 2094, South Africa; 0.03 mg/kg). Vatinoxan was diluted to 10 mg/ml and administered IM in the triceps muscle to each of the blesbok at 5 mg per milligram of medetomidine, 10 mg per milligram of medetomidine, and 20 mg per milligram of medetomidine, respectively, using a 21 gauge 1 in. needle. The results indicated that 5 and 10 mg of vatinoxan per milligram of medetomidine did not result in clinically significant changes in blood pressure, whereas 20 mg vatinoxan per milligram of medetomidine resulted in rapid drop in blood pressure and increase in heart rate. As a result, a vatinoxan dose of 15 mg/mg medetomidine was selected for the second phase of the study, which began after a washout period of 2 wk.

The second phase of the study, conducted in June and July 2022, consisted of a larger randomized, non–blinded cross–over study in which eight blesbok were immobilized on four separate occasions with a 2–wk washout period between each occasion.[31] During each occasion, the blesbok were allocated one of four immobilization protocols at random. For the purposes of this study, the results from only two of these protocols were analyzed: (1) thiafentanil—medetomidine (TM): 0.5 mg thiafentanil + 1.5 mg medetomidine + 1.5 ml saline and (2) thiafentanil—medetomidine + vatinoxan (TM + VAT): 0.5 mg thiafentanil + 1.5 mg medetomidine + 15 mg vatinoxan per milligram medetomidine (total 22.5 mg). The vatinoxan or saline were not administered in the dart but given IM at 10 min after recumbency.

The immobilizing drugs were administered via a CO_2 powered dart rifle (DanInject, 6000 Kolding, Denmark) with a 1.5–ml dart and a 1.5 × 25–mm collared needle (DanInject). The blesbok were not fed in the morning of the immobilization but water was available at all times except during the immobilization event. All animals were darted in the gluteal muscles and no animal received more than one dart. Ambient temperatures ranged from 7 to 24°C with a mean of 16.5°C. Time from injection to first signs of altered consciousness (time to first sign) and time from injection to recumbency (induction time) were recorded. Once recumbent, the blesbok was blindfolded, placed on a stretcher, and moved from the enclosure to a nearby shaded area for monitoring. The blesbok was maintained in sternal recumbency with the head elevated and the nose pointing down during the monitoring period. Either the auricular or the median artery of the metacarpus was catheterized with a 22–gauge Jelco® catheter (Midlands Veterinary Wholesalers, Germiston 1420, South Africa), which was closed with a yellow catheter stopper (In–stopper,

Midlands, Germiston 1420, South Africa). A portable blood pressure monitor (IntraTorr, United Kingdom, IntraVitals, https://www.intravitals.com) was connected to the catheter via a transducer (Deltran II pressure transducer, Utah Medical, Midvale, Utah 84047, USA) placed at heart level using non–compliant tubing (DPT–EL122, South African Hospital Supplies, Hout Bay 7872, South Africa) and 21 gauge × 1 in. needle inserted into the catheter stopper. The transducer was zeroed against atmospheric air before measurements began. The following variables were measured at 7, 10, 15, 20, 25, 30, and 35 min after recumbency: heart rate (HR), respiratory rate (RR), mean arterial blood pressure (MAP), peripheral hemoglobin oxygen saturation (SpO_2), rectal temperature (RT), and subjective assessment of the quality of immobilization as per the method described by Gaudio et al.[8] (Supplemental Table 1). SpO_2 was measured with a pulse oximeter with its reflectance probe fixed with tape to the skin under the tail (Nonin PalmSat 2500, 5026 RH Tilburg, Netherlands). Rectal temperature was measured by means of a digital thermometer (Hanna Checktemp 1, Hanna Instruments [Pty] Ltd, Woonsocket, Rhode Island 02895, USA). Respiratory rate was measured by visually counting the movement of the thorax and/or nares and heart rates by cardiac auscultation. Arterial blood samples (1 ml) were collected into a pre–heparinized 1–ml syringe at 10 min (just prior to the saline or vatinoxan administration), and 15, 25, and 35 min after recumbency. The catheter was flushed with sterile 0.9% saline after each blood collection. Blood gas analysis at 37°C (partial pressure of oxygen [PaO_2], partial pressure of carbon dioxide [$PaCO_2$], pH, and lactate [Lact]) was performed using a portable EPOC Blood Analysis Reader (Heska, Loveland, Colorado 80538, USA) using pre–calibrated EPOC BGEM smart cards (Epocal, Kyron Laboratories, Johannesburg 2094, South Africa) within 5 min after collection. The reader was also used to measure barometric pressure at the time of sampling as well as environmental temperature.

At the end of monitoring, the dart wound was treated, and the animal was weighed in the carry tarp using a hanging scale, carried back to its enclosure, and placed in sternal recumbency. The immobilizing effects of thiafentanil and medetomidine were antagonized with naltrexone (50 mg/ml, Wildlife Pharmaceuticals; 5 mg) and atipamezole (20 mg/ml, Kyron Laboratories; 7.5 mg) administered IM into the shoulder muscles. The time from injection to standing was recorded.

Data were summarized using descriptive statistics (Tables 1, 2) and the mean and standard deviation of each variable graphed by treatment group for each time point (Figs. 1, 2). The $P(A–a)O_2$ was calculated as $PAO_2 - PaO_2$, and PAO_2 was determined using the alveolar gas equation as previously described.[23] Significant differences in physiological variables between the two treatment groups was assessed using two–way repeated–measures analysis of variance using the R package rstatix.[18] Differences at each time point were assessed using the Bonferroni post hoc test. Graphs were created using the R package ggplot.[35]

RESULTS

All blesbok were immobilized with a mean dose of 0.01 mg/kg (SD = 0.001 mg/kg; range = 0.008–0.010 mg/kg) thiafentanil and 0.03 mg/kg (SD = 0.002 mg/kg, range = 0.024–0.032 mg/kg) medetomidine. The mean time from injection to first sign was 5 min (SD = 3 min; range = 2–15 min). The mean induction time (from darting to recumbency) averaged 12:53 min (SD 5:28 min; range = 6:00 – 27:21 min). Recovery time (from reversal to standing) was not significantly different between treatments and averaged 10:26 min (SD = 1:56; range = 8:15 – 13:25 min) for the TM and 11:05 min (SD = 3:20; range = 7:00 – 15:49) for the TM + VAT group. Heart rates were <40 beats/min in all but one blesbok at the 7– and 10–min time points (Table 1). There was a significant interaction between treatment and time ($F = 6.96$, $P = 0.008$), and after administration of vatinoxan, the HR were significantly higher at the 15 ($P = 0.039$), 20 ($P = 0.029$), and 25 ($P = 0.028$) min time points compared to when no vatinoxan was administered (Table 1, Fig. 1).

Respiratory rates significantly increased over time for both the TM and TM + VAT treatments ($F = 5.27$, $P = 0.03$). The RR was significantly higher in the TM group at 7 min post–recumbency compared to the TM + VAT group ($P = 0.04$); however, that was before vatinoxan was administered and therefore not related to treatment (Table 1, Fig. 1). The MAP significantly decreased over time for both treatments ($F = 33.06$, $P = 0.001$), and there was an interaction between time and treatment ($F = 38.65$, $P = 0.001$), with the MAP being significantly lower in the TM + VAT group at the 25 ($P = 0.034$), 30 ($P = 0.012$) and 35 ($P = 0.011$) min time points compared to the TM group (Table 1, Fig. 1). Rectal temperature, SpO_2, PaO_2, and $PaCO_2$ did not significantly differ between the two treatments or at any time points (Table 2, Fig. 2). The $P(A–a)O_2$

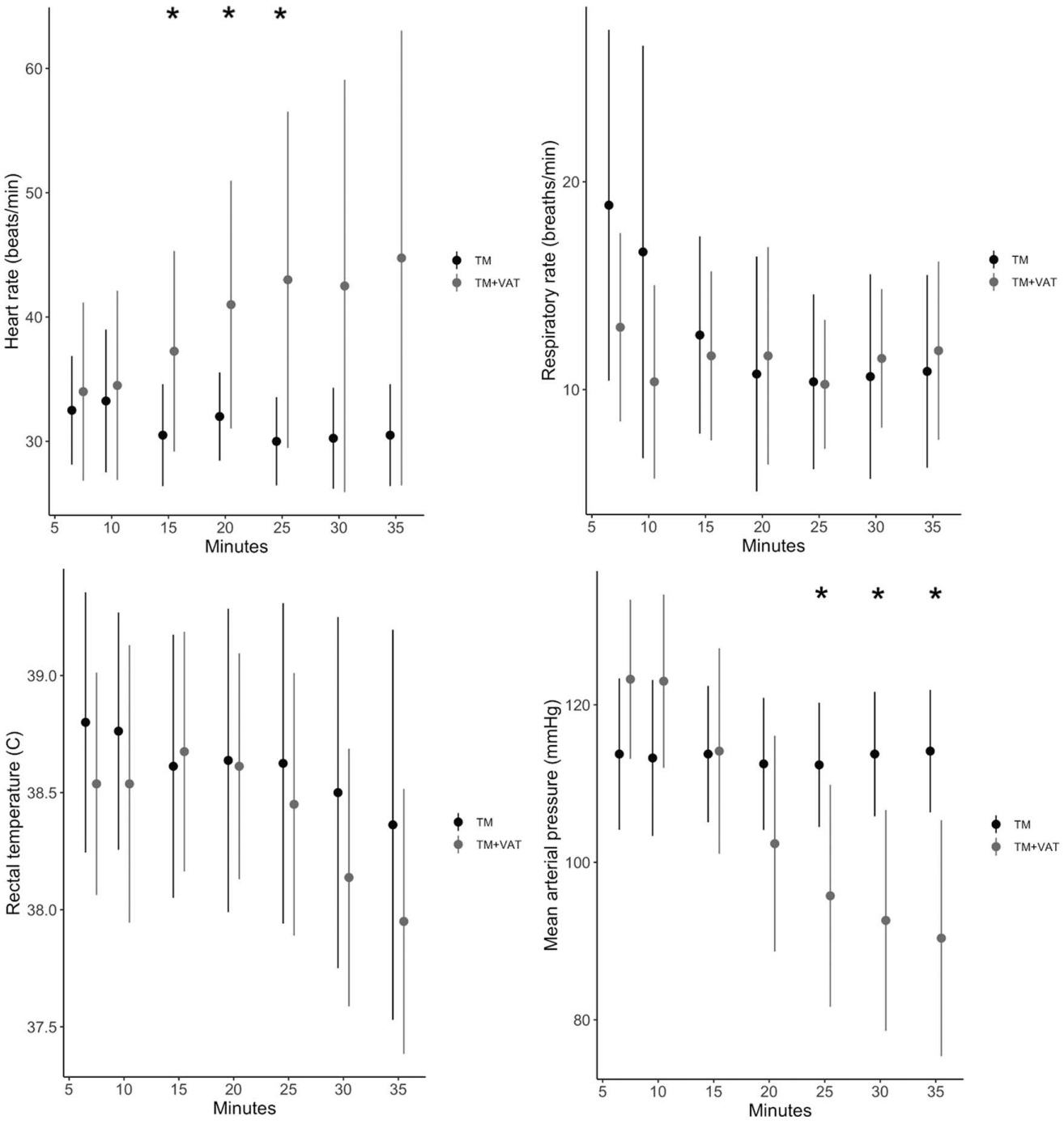

Figure 1. Mean and standard deviation of the heart rate (beats per minute), respiratory rate breaths per minute), rectal temperature (°C), and mean arterial blood pressure (mmHg) in eight adult female blesbok immobilized twice with 0.5 mg thiafentanil and 1.5 mg medetomidine. Either saline or 22.5 mg vatinoxan was injected intramuscularly immediately after the 10min sampling time point in each treatment (thiafentanil–medetomidine [TM] and thiafentanil–medetomidine + vatinoxan [TM + VAT]), respectively. The asterisk indicates a significant difference between TM and TM + VAT at the time point.

gradient significantly decreased over time for both treatments ($F = 13.25$, $P = 0.003$), and was significantly lower in the TM group compared to the TM + VAT–treated animals at the 10–min time point ($P = 0.03$); however, that was before vatinoxan was administered and was therefore not related to the vatinoxan administration (Table 2, Fig. 2). The sedation level was adequate for handling for all blesbok and classified as sedation score = 3 (light; Supplementary Table 1) for two of eight blesbok in both treatment groups at the 7– and 10–min time points, and light for one of eight blesbok in the vatinoxan group at the 15–25–min time point (Table 1). For the remainder of the blesbok the sedation level was classified as sedation score = 4 (deep; Table 1). The pH was

Table 1. Sedation score[8] and mean physiological variables (±SD; range) recorded during immobilization of eight adult, female blesbok with TM (thiafentanil + medetomidine) and TM with vatinoxan (TM + VAT) injected immediately after the 10–min sample collection.

Time (min)	Treatment	Sedation score	HR (beats/minute)	RR (breaths/minute)	RT (°C)	MAP (mmHg)	SpO_2 (%)
7	TM	3 ($n = 2$), 4 ($n = 6$)	33 ± 4.4; 28–40	19 ± 8.4; 8–30	38.8 ± 0.6; 37.9–39.7	114 ± 9.6; 98–125	88 ± 11.9; 67–98
	TM	3 ($n = 2$), 4 ($n = 6$)	34 ± 7.2; 28–50	13 ± 4.5; 8–20	38.5 ± 0.5; 37.7–38.9	123 ± 10.1; 111–144	93 ± 6.7; 80–100
10	TM	3 ($n = 2$), 4 ($n = 6$)	33 ± 5.8; 24–40	17 ± 9.9; 7–30	38.8 ± 0.5; 37.9–39.4	113 ± 9.9; 100–125	90 ± 9.8; 71–98
	TM (VAT inj)	3 ($n = 2$), 4 ($n = 6$)	35 ± 7.6; 28–50	10 ± 4.7; 4–18	38.5 ± 0.6; 37.5–39.1	123 ± 11.0; 111–144	91 ± 5.9; 85–99
15	TM	3 ($n = 1$), 4 ($n = 7$)	31[a] ± 4.1; 24–36	13 ± 4.7; 5–20	38.6 ± 0.6; 37.8–39.3	114 ± 8.7; 101–123	90 ± 10.2; 68–100
	TM + VAT	3 ($n = 1$), 4 ($n = 7$)	37 ± 8.1; 28–50	12 ± 4.1; 6–18	38.7 ± 0.5; 37.8–39.2	114 ± 13.0; 95–136	89 ± 6.0; 82–96
20	TM	4 ($n = 8$)	32[a] ± 3.5; 28–36	11 ± 5.7; 4–20	38.6 ± 0.6; 37.6–39.7	113 ± 8.4; 101–123	92 ± 6.2; 83–100
	TM + VAT	3 ($n = 1$), 4 ($n = 7$)	41 ± 9.9; 28–60	12 ± 5.2; 4–20	38.6 ± 0.5; 37.9–39.2	102[b] ± 13.7; 83–127	89 ± 6.2; 78–96
25	TM	4 ($n = 8$)	30[a] ± 3.5; 24–36	10 ± 4.2; 4–16	38.6 ± 0.7; 37.5–39.8	112[a] ± 7.9; 102–123	91 ± 9.1; 73–100
	TM + VAT	3 ($n = 1$), 4 ($n = 7$)	43 ± 13.5; 26–70	10 ± 3.1; 6–14	38.5 ± 0.6; 37.8–39.4	96[b] ± 14.1; 77–121	93 ± 5.0; 83–100
30	TM	4 ($n = 8$)	30 ± 4.1; 24–36	11 ± 4.9; 4–18	38.5 ± 0.8; 37.3–39.9	114[a] ± 7.9; 103–123	92 ± 5.9; 82–99
	TM + VAT	4 ($n = 8$)	43 ± 16.6; 24–78	12 ± 3.3; 8–16	38.1[b] ± 0.6; 37.4–38.8	93 ± 14.0; 74–115	94 ± 4.9; 85–99
35	TM	4 ($n = 8$)	31 ± 4.1; 24–36	11 ± 4.6; 4–16	38.4 ± 0.8; 37.0–39.9	114[a] ± 7.8; 102–123	93 ± 4.7; 87–99
	TM + VAT	4 ($n = 8$)	45 ± 18.3; 26–84	12 ± 4.3; 7–18	37.9[b] ± 0.6; 37.2–38.6	90 ± 14.9; 71–115	93 ± 4.7; 85–99

[a] Significant differences ($P \leq 0.05$) between TM and TM + VAT.
[b] Significant difference ($P \leq 0.05$) from previous time point within the same treatment.

significantly lower in the TM + VAT group at 25 min, but not at any other time point (Table 2). There was no significant difference in lactate at any time point and lactate concentrations were low (<1.4 mmol/L; Table 2) in all animals.

DISCUSSION

The adult, female blesbok were effectively immobilized with 0.5 mg of thiafentanil and 1.5 mg of medetomidine; however, induction times varied between animals. A consistently rapid induction is critical when immobilizing free–ranging animals, and this combination may therefore be most suitable for boma–confined or calm animals.

The majority of blesbok had heart rates <40 bpm at the 7– and 10–min time points, and the mean HR of the TM animals remained <40 bpm for the entire immobilization period. Heart rates of 58–97 bpm are considered normal in conscious blesbok,[5] and the combination therefore induced bradycardia, which improved after administration of vatinoxan. Normal MAP in blesbok is unknown, but for goats a MAP of 70–110 mmHg is considered normal[10] and <65 mmHg consistent with hypotension in anesthetized goats.[3] The MAP was therefore mildly elevated in all blesbok, and intramuscular administration of vatinoxan at 15 mg vatinoxan per milligram of medetomidine reduced the MAP to within the normal blood pressure range for the majority of the blesbok. Vatinoxan antagonizes the peripheral vasoconstriction induced by the $alpha_2$–agonists while having very limited central effect, because of low lipophilia and poor penetration of the blood brain barrier.[4] When the vasoconstriction is antagonized, there is a compensatory increase in heart rate to maintain normotension.[12,28,30] The blood pressure was only moderately elevated to begin with, which likely explains the relatively small increase in HR after vatinoxan administration in the blesbok. Vatinoxan may improve peripheral tissue perfusion by antagonizing the vasoconstrictive effect of the $alpha_2$–agonists as shown in dogs and horses;[24,25] however, further confirmation of these effects is required.

The cardiovascular effects of $alpha_2$–agonists also influences gas exchange in the lungs through ventilation–perfusion mismatching.[22,29] The exact pulmonary effects of $alpha_2$–agonists in blesbok are unknown, but in domestic sheep they cause pulmonary edema shortly after administration.[19] Potent opioids can also induce pulmonary hypertension,[23] and the resulting pulmonary congestion, possibly edema, and reduction in capillary blood transit time may further hamper effective gas exchange.[23,26] Hypoxemia is generally defined as a $PaO_2 < 80$ mmHg and an $SpO_2 < 95\%$, and severe hypoxemia

Table 2. Mean arterial blood gases (±SD; range) and $P(A–a)O_2$ gradient recorded during immobilization of eight adult, female blesbok with TM (thiafentanil + medetomidine) and TM with vatinoxan (VAT) injected immediately after the 10–min sample collection.

Time (min)	Treatment	PaO_2 (mmHg)	$PaCO_2$ (mmHg)	$P(A–a)O_2$ gradient (mmHg)	pH	Lactate (mmol/L)
10	TM	57.4 ± 7.4; 45.9–68.5	38.7 ± 2.4; 34.0–41.4	40.3 ± 6.9; 29.8–51.7	7.48 ± 0.03; 7.44–7.53	0.81 ± 0.32; 0.46–1.39
	TM (VAT inj)	51.4 ± 7.5; 38.8–61.1	38.5 ± 5.0; 31.2–46.8	48.3 ± 5.5; 39.7–55.2	7.48 ± 0.01; 7.40–7.54	0.83 ± 0.28; 0.48–1.27
15	TM	54.6 ± 6.6; 44.1–61.9	40.5 ± 4.1; 34.0–46.6	41.4 ± 5.7; 34.5–52.7	7.45 ± 0.02; 7.40–7.49	0.79 ± 0.29; 0.47–1.32
	TM + VAT	50.8 ± 7.0; 41.0–61.0	40.4 ± 4.7; 33.9–47.6	46.9 ± 4.7; 40.4–54.4	7.45 ± 0.02; 7.42–7.81	0.83 ± 0.26; 0.52–1.20
25	TM	54.6 ± 8.2; 41.9–68.4	43.5[b] ± 3.3; 38.7–47.6	38.3 ± 7.8; 25.2–52.2	7.44[a] ± 0.02; 7.40–7.47	0.77 ± 0.24; 0.47–1.22
	TM + VAT	50.4 ± 6.7; 41.9–58.5	46.3[b] ± 4.7; 40.4–52.6	41.7 ± 6.4; 31.4–48.9	7.40[a] ± 0.02; 7.36–7.44	0.75 ± 0.22; 0.43–1.03
35	TM	56.1 ± 7.1; 43.3–65.3	45.3 ± 7.3; 38.9–61.8	35.2 ± 9.8; 16.4–48.6	7.43 ± 0.04; 7.33–7.47	0.77 ± 0.22; 0.52–1.18
	TM + VAT	56.8 ± 6.3; 42.3–63.8	46.8 ± 6.4; 38.6 – 54.7	34.9 ± 6.3; 25.7–42.0	7.40 ± 0.03; 7.37–7.44	0.75 ± 0.17; 0.51–1.02

[a] Significant differences ($P \leq 0.05$) between TM and VAT.
[b] Significant difference ($P \leq 0.05$) from previous time point within the same treatment.

is defined as a $PaO_2 < 60$ mmHg and an SpO_2 of less than 90%.[11] All blesbok were therefore severely hypoxemic on this combination. The impaired gas exchange in the lungs was reflected by the elevated $P(A–a)O_2$ gradients. An elevated gradient indicates suboptimal alveolar–arteriolar oxygen transfer, which can be caused by pulmonary hypertension, resulting in congestion and alveolar–interstitial edema, or ventilation/perfusion mismatching.[20] A reference value for normal $P(A–a)O_2$ gradients is not available for blesbok, but in small ruminants, a gradient of 20–25 mmHg is considered normal.[23] Because of the $P(A–a)O_2$ gradient decreased in both the TM and TM + VAT group over time, the administration of vatinoxan did not lead to a statistically significant improvement in the gas exchange when comparing to the $P(A–a)O_2$ gradients in the TM animals.

Potent opioids may also cause centrally mediated respiratory depression, which can reduce ventilation and riddance of CO_2.[23] All blesbok had respiratory rates that were slower than proposed normal respiratory rates for resting blesbok of 13–17 breaths/min.[5] In small ruminants, the $PaCO_2$ is expected to be 40 mmHg +/− 7 mmHg.[15] The $PaCO_2$ values measured were not severely elevated in any animal, and improved over the course of the immobilization, indicating adequate ventilation and elimination of CO_2 for most animals. In sum, there was no significant difference in the SpO_2, PaO_2, and $P(A–a)O_2$ gradient between the TM and TM + VAT animals indicating that the vatinoxan did not induce a measurable improvement in the gas exchange in the lungs. Our findings are consistent with studies of red deer and wild boars anesthetized with medetomidine–tiletamine–zolazepam that reported no significant difference in SpO_2 (red deer) or PaO_2 (wild boar) after vatinoxan administration.[6,7] Whether an improvement in oxygenation would have been observed if vatinoxan could have been co–administered with the thiafentanil and medetomidine in the dart requires further investigation. In sevoflurane–anesthetized sheep, administration of vatinoxan prior to injection of dexmedetomidine prevented dexmedetomidine–induced hypoxemia, bronchoconstriction, and edema.[1] However, vatinoxan is not very soluble, so the co–administration with the immobilization drugs would not have been possible without the use of large darts, which is neither practical nor ethical in small antelope species such as blesbok.

Our data are limited by the small sample size. Although all blesbok were healthy throughout the study, some observations may have been influenced more by individual characteristics of specific study

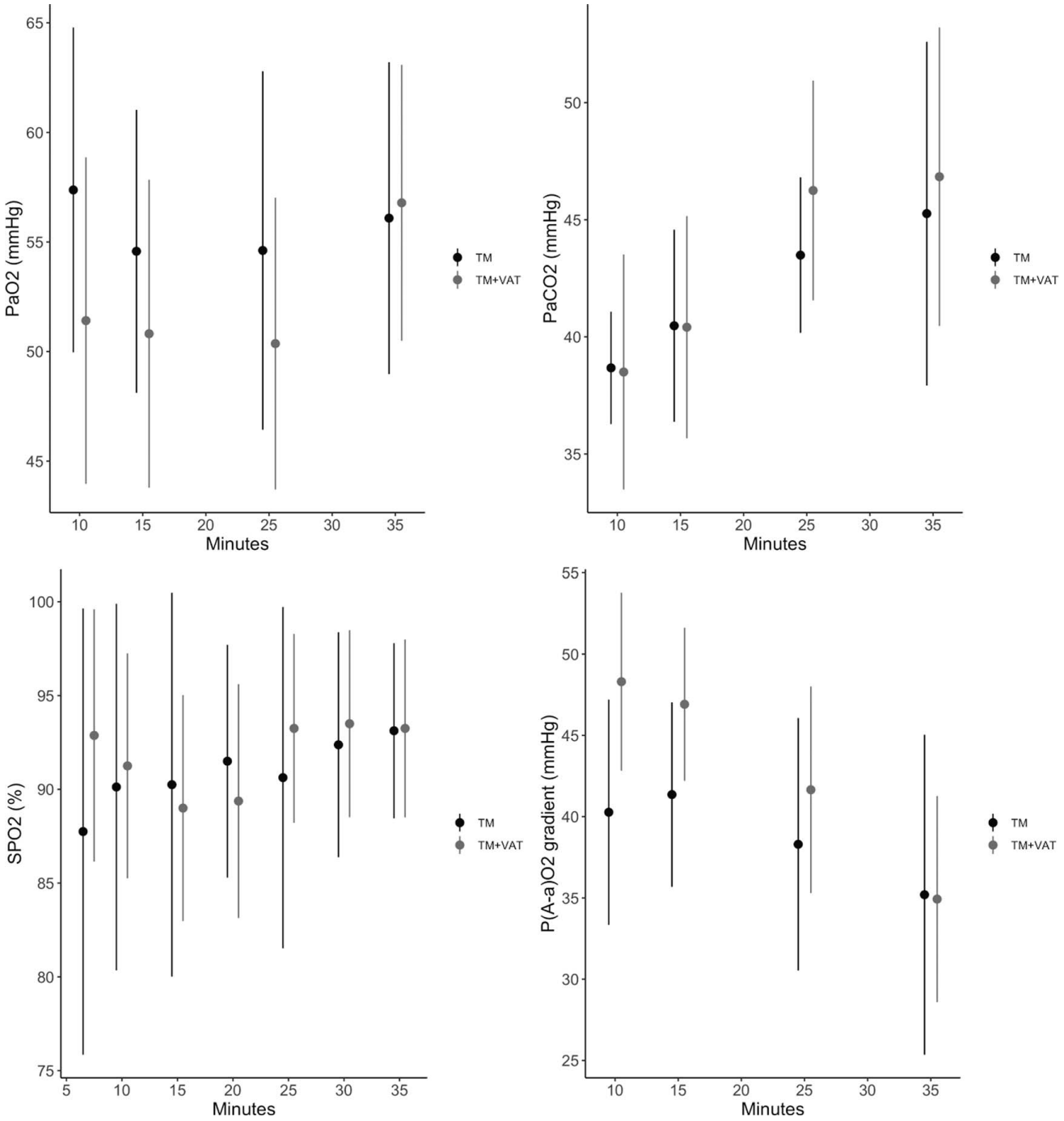

Figure 2. Mean and standard deviation of the PaO_2, $PaCO_2$, SpO_2, and $P(A–a)O_2$ gradient in eight adult female blesbok immobilized twice with 0.5 mg thiafentanil and 1.5 mg medetomidine. Either saline or 22.5 mg vatinoxan was injected intramuscularly immediately after the 10–min sampling time point in each treatment (thiafentanil–medetomidine [TM] and thiafentanil–medetomidine + vatinoxan [TM + VAT]), respectively.

animals, and it is unknown whether there was any carryover effect from the repeat immobilizations.

CONCLUSION

In conclusion, blesbok were effectively immobilized with 0.5 mg of thiafentanil and 1.5 mg of medetomidine. Induction times ranged widely. All blesbok experienced bradycardia, hypoxemia, respiratory depression, and mild hypertension. The administration of vatinoxan attenuated hypertension in most animals and increased the heart rate. There was no significant difference in the SpO_2, PaO_2, $PaCO_2$, or $P(A–a)O_2$ gradients between treatments. The administration of vatinoxan did therefore not lead to any significant or clinically relevant improvement of the severe hypoxemia that these blesbok experienced on this TM

combination and oxygen administration is recommended. The change in cardiovascular variables may suggest that tissue perfusion is improved with vatinoxan administration, positive outcomes that require further investigation.

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Conflict of interest statement: JPR has a financial interest in Wildlife Pharmaceuticals, and LLL., also a co–author, is employed by Wildlife Pharmaceuticals. Wildlife Pharmaceuticals contributed towards this project financially. However, we do not believe that this would have inappropriately influenced the analysis or writing of this article.

LITERATURE CITED

1. Adam M, Huuskonen V, Raekallio MR, Casoni D, Mykkänen AK, Lappalainen AK, Kajula M, Kallio–Kujala IJ, Vainio OM. Cardiopulmonary effects of vatinoxan in sevoflurane–anaesthetised sheep receiving dexmedetomidine. Vet J. 2018:238(1):63–69.

2. Adam M, Lindén J, Raekallio M, Meller A, Mannerström B, Abu–Shahba A, Seppänen–Kaijansinkko R, Salla K. Effects of vatinoxan on xylazine–induced pulmonary alterations in sheep. J Vet Pharmacol Ther. 2022;45(1):117–125.

3. Clarke KW, Trim CM, Hall LW. Veterinary anaesthesia. 11th ed. St. Louis (MO): Elsevier Saunders; 2014.

4. Clineschmidt BV, Pettibone DJ, Lotti VJ, Hucker HB, Sweeney BM, Reiss DR, Lis EV, Huff JR, Vacca J. A peripherally acting alpha–2 adrenoceptor antagonist: L–659,066. J Pharmacol Exp Ther. 1988;245(1):32–40.

5. Du Plessis D. The potential of midazolam for use as sedative for blesbok (*Damaluscus pygargus phillipsi*). Thesis (MScAgric); 2018. Stellenbosch University (South Africa).

6. Einwaller J, Meyer LCR, Auer U, Raekallio M, Nowack J, Haw A, Vetter S, Painer J, Stalder G. Cardiovascular effects of intravenous vatinoxan in wild boars (*Sus scrofa*) anaesthetised with intramuscular medetomidine-tiletamine-zolazepam. Vet Rec. 2021;190(2):e835.

7. Einwaller J, Painera J, Raekallio M, Gascha K, Restituttic F, Auerc U, Stalder GL. Cardiovascular effects of intravenous vatinoxan (MK–467) in medetomidine–tiletamine–zolazepam anaesthetised red deer (*Cervus elaphus*). Vet Anaesth Analg. 2020;47(4):518–527.

8. Gaudio E, Laubscher LL, Pfitzer S, Raath JP, Hoffman LC, De Benedictis GM. Immobilization quality and cardiopulmonary effects of etorphine alone compared with etorphine–azaperone in blesbok (*Damaliscus pygargus phillipsi*). Vet Anaesth Analg. 2020;47(4):528–536.

9. Greunz EM, Limón D, Petersen RL, Raekallio MR, Grøndahl C, Bertelsen MF. The impact of vatinoxan on medetomidine–ketamine–midazolam immobilization in Patagonian maras (*Dolichotis patagonum*). Vet Anaesth Analg. 2021;48(3):372–379.

10. Grimm KA, Lamont LA, Tranquilli WJ, Greene SA, Robertson SA. Veterinary anesthesia and analgesia—Lumb and Jones. 5th ed. Ames (IA): Wiley–Blackwell; 2015.

11. Haskins SC. Chapter 15 – Hypoxemia. In Silverstein DC, Hopper K (eds.). Small animal critical care medicine. 2nd ed. St. Louis (MO): W.B. Saunders; 2015. p. 81–86.

12. Honkavaara JM, Raekallio MR, Syrja PM, Pypendop BH, Knych HK, Kallio–Kujala IJ, Vainio OM. Concentrations of medetomidine enantiomers and vatinoxan, an α_2–adrenoceptor antagonist, in plasma and central nervous tissue after intravenous coadministration in dogs. Vet Anaesth Analg. 2020;47(1):47–52.

13. Honkavaara JM, Restitutti F, Raekallio MR, Kuusela EK, Vainio OM. The effects of increasing doses of MK–467, a peripheral alpha(2)–adrenergic receptor antagonist, on the cardiopulmonary effects of intravenous dexmedetomidine in conscious dogs. J Vet Pharmacol Ther. 2011;34(4):332–337.

14. Hoque A. Various systemic effects of MK–467, a peripherally acting α2–adrenoceptor antagonist in different animal species: an overview. Agric Rev. 42(3): 348–352.

15. Ismail ZB, Jawasreh K, Al–Majali A. Effect of xylazine–ketamine–diazepam anesthesia on certain clinical and arterial blood gas parameters in sheep and goats. Comp Clin Pathol. 2010;19:11–14. doi:10.1007/s00580–009–0896–6

16. Jaeger AT, Pypendop BH, Ahokoivu H, Honkavaara J. Cardiopulmonary effects of dexmedetomidine, with and without vatinoxan, in isoflurane–anesthetized cats. Vet Anaesth Analg. 2019;46(6):753–764.

17. Jalanka HH, Roeken BO. The use of medetomidine, medetomidine-ketamine combinations, and atipamezole in nondomestic mammals: a review. J Zoo Wildl Med. 1990;21(3):259–282.

18. Kassambara A. Rstatix: Pipe–friendly framework for basic statistical tests. 2021. R Package Rstatix Version 0.7.0. https://CRAN.R–project.org/package=rstatix

19. Kästner SBR. Alpha2–agonists in sheep: a review. Vet Anaesth Analg. 2006;33(2):79–96.

20. Kock MD, Burroughs R. Chemical and physical restraint of African wild animals. 3rd ed. Greyton (South Africa): International Wildlife Veterinary Services (Africa); 2021.

21. Kumar R, Singh M, Verma NK, Akash, Sasikala R, Kumar R, Hoque M. Systemic effects and

clinical application of medetomidine in animals: a review. Indian J Anim Res. 2020;54(9):1063–1068.

22. Laubscher LL, Meyer LCR, Laurence M, Raath JP. A comparison of immobilisation quality and cardiorespiratory effects of etorphine-azaperone versus etorphine-midazolam combinations in blesbok. J S Afr Vet Assoc. 2022;93(1):1–9.

23. Meyer LCR, Hetem RS, Mitchell D, Fuller A. Hypoxia following etorphine administration in goats (*Capra hircus*) results more from pulmonary hypertension than from hypoventilation. BMC Vet Res. 2015;11(1): 1–9.

24. Neudeck S, Twele L, Kopp V, Kästner S. Pharmacodynamics and plasma concentrations of dexmedetomidine with or without vatinoxan as a constant-rate infusion in horses anaesthetized with isoflurane—a pilot study. J Vet Pharmacol Ther. 2021;44(5):754–765.

25. Niemann L, Kutter APN, Joerger FB, Wieser ML, Hartnack S, Steblaj B. The impact of vatinoxan on microcirculation after intramuscular co-administration with medetomidine in Beagle dogs: a blinded crossover study. Vet Anaesth Analg. 2022;49(4):336–343.

26. Pfitzer S, Laurence M, Laubscher LL, Raath JP, Warren K, Vaughen–Higgins R, Meyer LCR. Do potent immobilising–opioids induce different physiological effects in impala and blesbok? J S Afr Vet Assoc. 2020;91(1):1–8.

27. Posner LP. Chapter 14. Sedatives and tranquilizers. In Riviere JE, Papich MG (eds.). Veterinary pharmacology and therapeutics. Hoboken (NJ): Wiley; 2018. p. 324–368.

28. Raekallio MR, Honkavaara JM, Vainio OM. The effects of L–659,066, a peripheral α2–adrenoceptor antagonist, and verapamil on the cardiovascular influences of dexmedetomidine in conscious sheep. J Vet Pharmacol Ther. 2010;33(5):434–438.

29. Read MR. A review of alpha2 adrenoreceptor agonists and the development of hypoxemia in domestic and wild ruminants. J Zoo Wildl Med. 2003;34(2): 134–138.

30. Restitutti F, Honkavaara JM, Raekallio MR, Kuusela EK, Vainio OM. Effects of different doses of L–659'066 on the bispectral index and clinical sedation in dogs treated with dexmedetomidine. Vet Anaesth Analg. 2011;38(5):415–422.

31. Roug A, Smith C, Raath J, Meyer L, Laubscher L. Efficacy and safety of three different opioid–based immobilisation combinations in blesbok (*Damaliscus pygargus phillipsi*). J S Afr Vet Assoc. 2023;94:145–152.

32. Sainmaa S, Mykkänen A, Adam M, Jantunen N, Vainio O, Raekallio M. Intravenous vatinoxan in markhors (*Capra falconeri heptneri*) immobilized with intramuscular medetomidine and ketamine—a preliminary dose screening study. J Zoo Wildl Med. 2019;50(1):159–166.

33. Tapio H. Impact of vatinoxan on cardiopulmonary and gastrointestinal effects of medetomidine and detomidine in horses. Dissertation; 2020. University of Helsinki (Finland).

34. West G, Heard D, Caulkett N. Zoo animal and wildlife immobilization and anaesthesia. Ames (IA): Blackwell; 2007.

35. Wickham H. ggplot2: Elegant graphics for data analysis. 2nd ed. New York (NY): Springer; 2016.

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